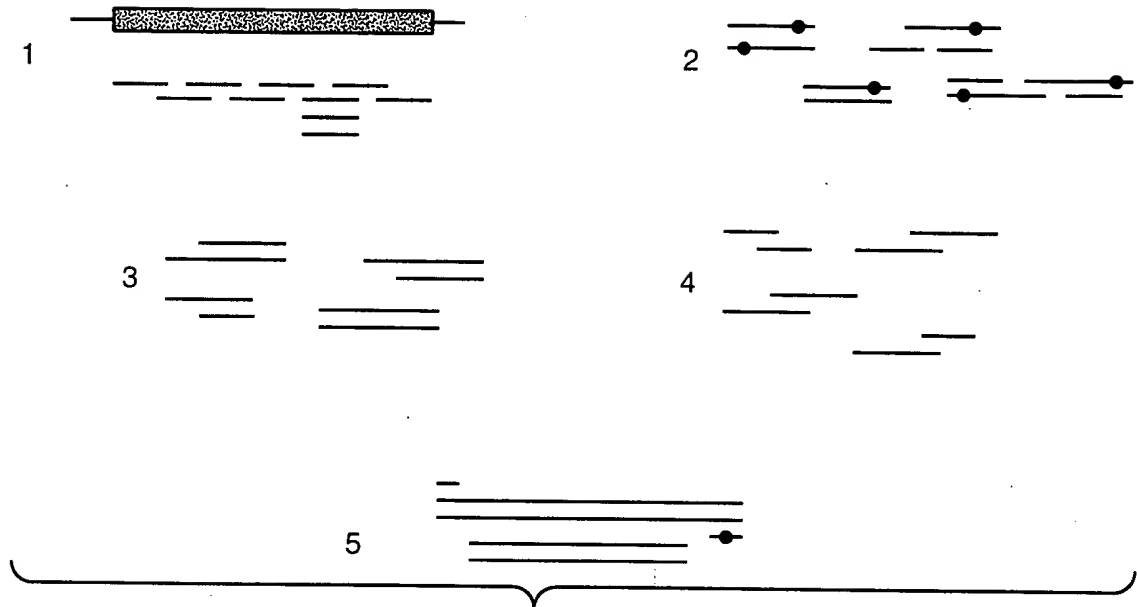
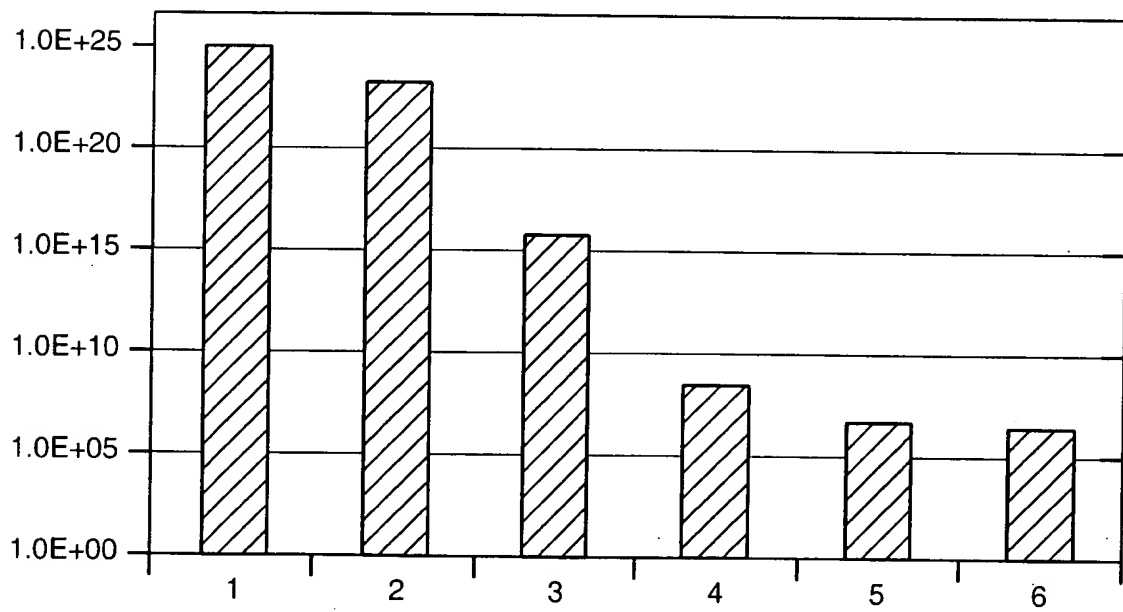


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**FIG. 1**



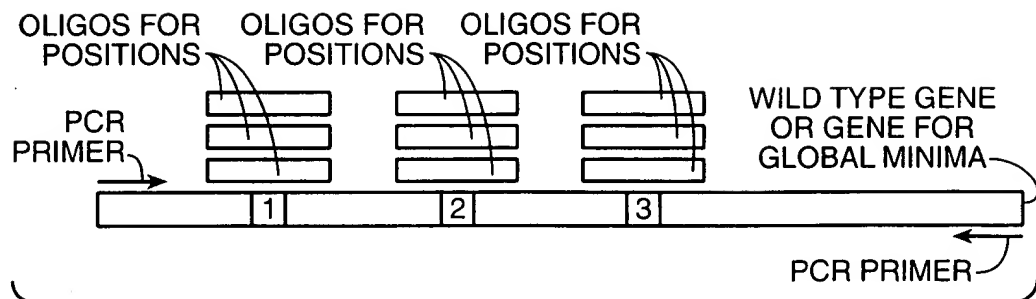
**FIG. 2**

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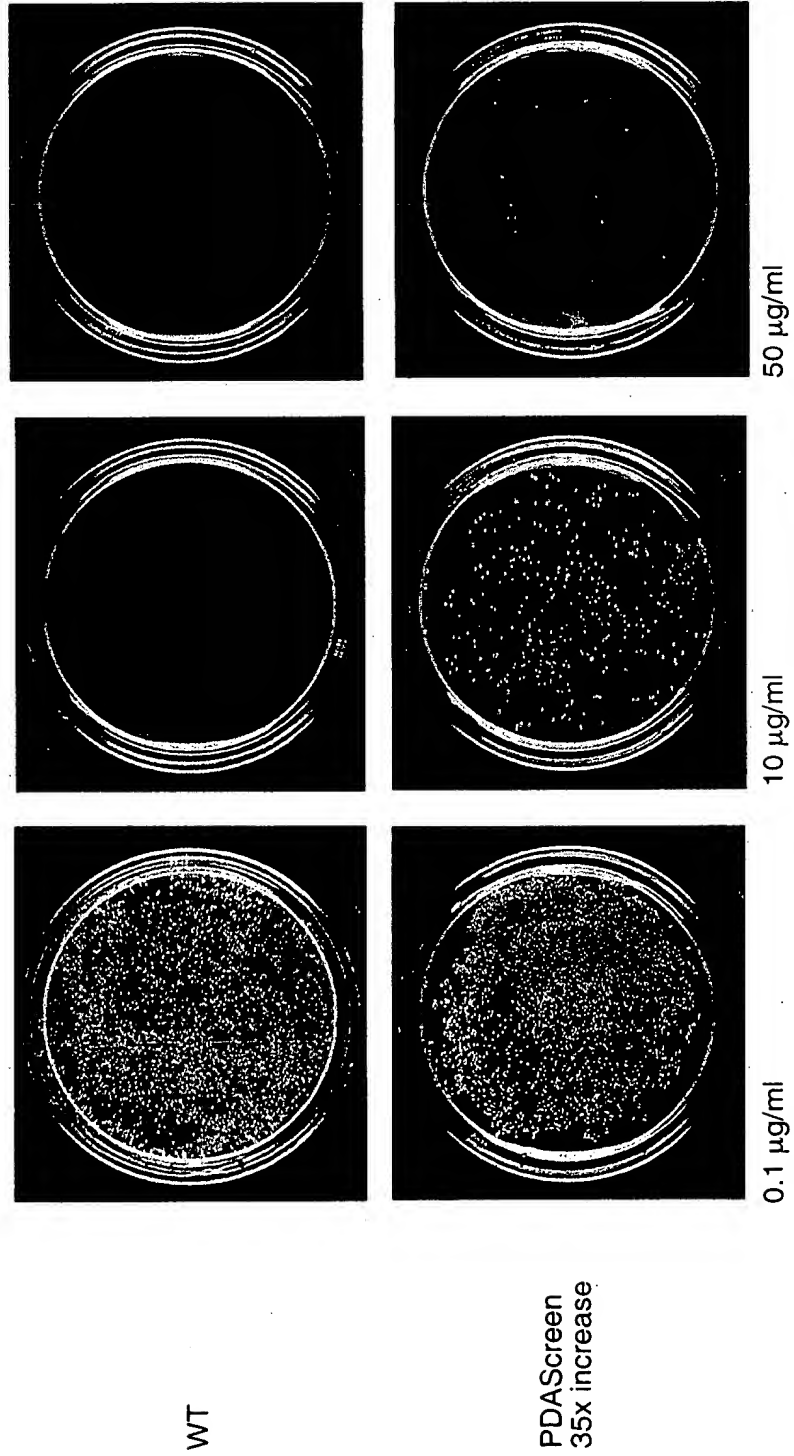
**FIG.\_3**



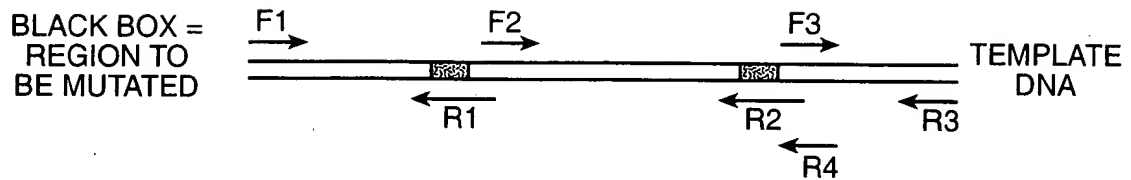
**FIG.\_5**

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**FIG. 4**



STEP 1: SET UP 3 PCR REACTIONS:

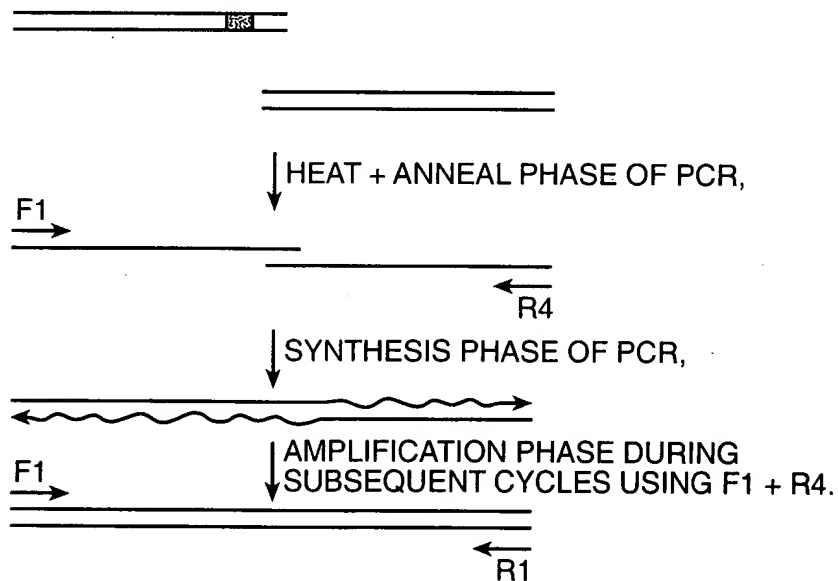
PRODUCTS:

TUBE 1:

TUBE 2:

TUBE 3:

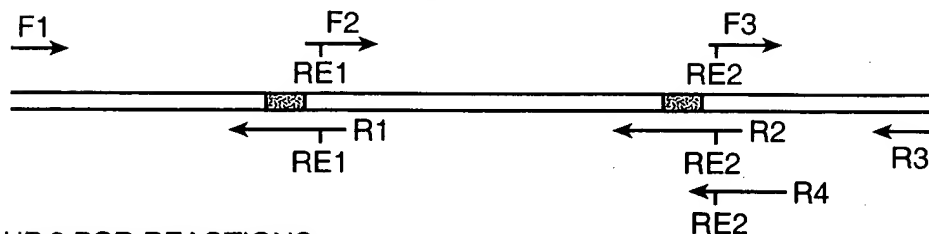
STEP 2: SET UP PCR REACTION WITH PRODUCTS OF TUBE 1 + PRODUCTS TUBE 2 + F1 + R4.



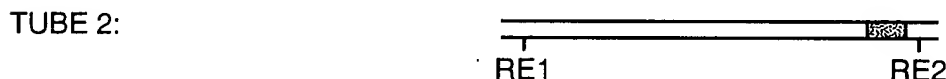
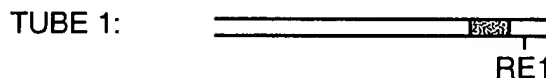
STEP 3: REPEAT STEP 2 USING PRODUCT FROM STEP 2 + PRODUCT FROM STEP 1, TUBE 3 + PRIMERS F1 + R3.

**FIG.\_6**

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**STEP 1:** SET UP 3 PCR REACTIONS:



**STEP 2:** DIGEST PRODUCTS FROM STEP 1 WITH SUITABLE RESTRICTION ENDONUCLEASES.

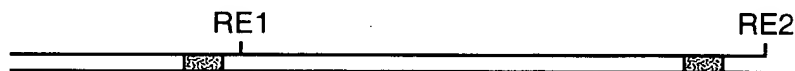
**STEP 3:** LIGATE DIGESTED PRODUCT FROM STEP 2, TUBE 2 WITH DIGESTED PRODUCT FROM STEP 2, TUBE 1.



**STEP 4:** AMPLIFY VIA PCR LIGATED PRODUCTS OF STEP 3 WITH F1 + R4.



**STEP 5:** DIGEST AMPLIFIED PRODUCT OF STEP 4 WITH RESTRICTION ENDONUCLEASE #2.



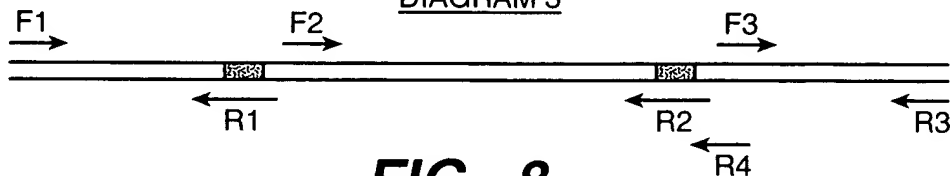
**STEP 6:** LIGATE PRODUCT FROM STEP 5 WITH PRODUCT FROM STEP 2, TUBE 1.



**STEP 7:** AMPLIFY PRODUCT FROM STEP 6 WITH F1 + R3.

**FIG. 7**

**DIAGRAM 3**



**FIG. 8**

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Amplification Scheme Based on M13 Single Stranded Template

Amplification Scheme & Math

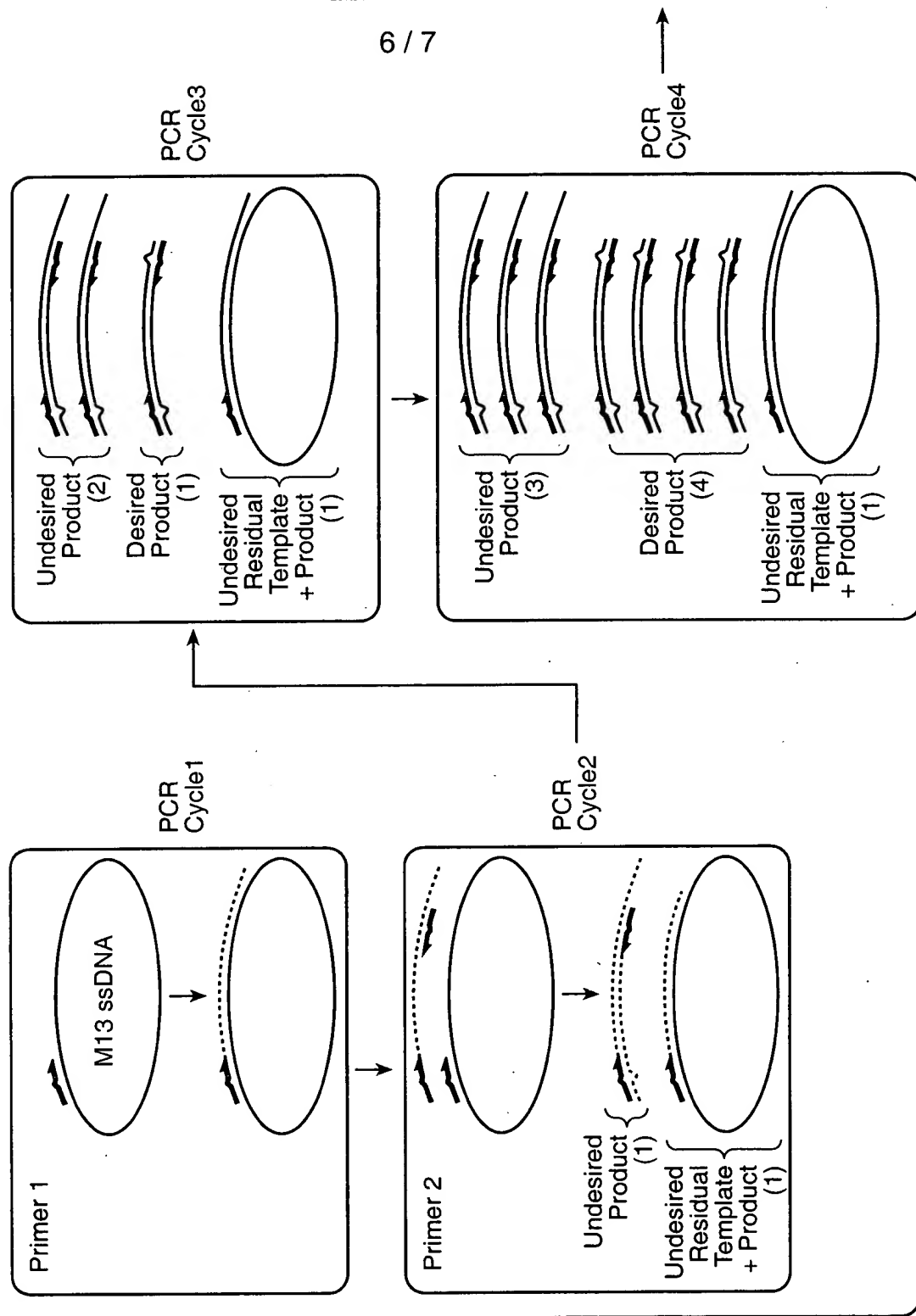


FIG. 9A

FOOTPRINT 06/2660

### Amplification Scheme Based on M13 Single Stranded Template

Numerical Progression of Desired Product  
with Increasing PCR Cycles

| PCR<br>Cycles | Desired<br>Product | Undesired<br>Products and<br>Residual<br>Template | Percent Desired<br>Product in Total<br>Product |
|---------------|--------------------|---|--|
| 1             |                    | 1   |  |
| 2             | 0                  | 2   | 0.00%  |
| 3             | 1                  | 3   | 25.00%   |
| 4             | 4                  | 4   | 50.00%   |
| 5             | 11                 | 5   | 68.75%   |
| 6             | 26                 | 6   | 81.25%   |
| 7             | 57                 | 7   | 89.06%   |
| 8             | 120                | 8   | 93.75%   |
| 9             | 247                | 9   | 96.48%   |
| 10            | 502                | 10  | 98.05%   |
| 11            | 1013               | 11  | 98.93%   |
| 12            | 2036               | 12  | 99.41%   |
| 13            | 4083               | 13  | 99.68%   |
| 14            | 8178               | 14  | 99.83%   |
| 15            | 16369              | 15  | 99.91%   |
| 16            | 32752              | 16  | 99.95%   |
| 17            | 65519              | 17  | 99.97%   |
| 18            | 131054             | 18  | 99.99%   |
| 19            | 262125             | 19  | 99.99%   |
| 20            | 524268             | 20  | 100.00%  |

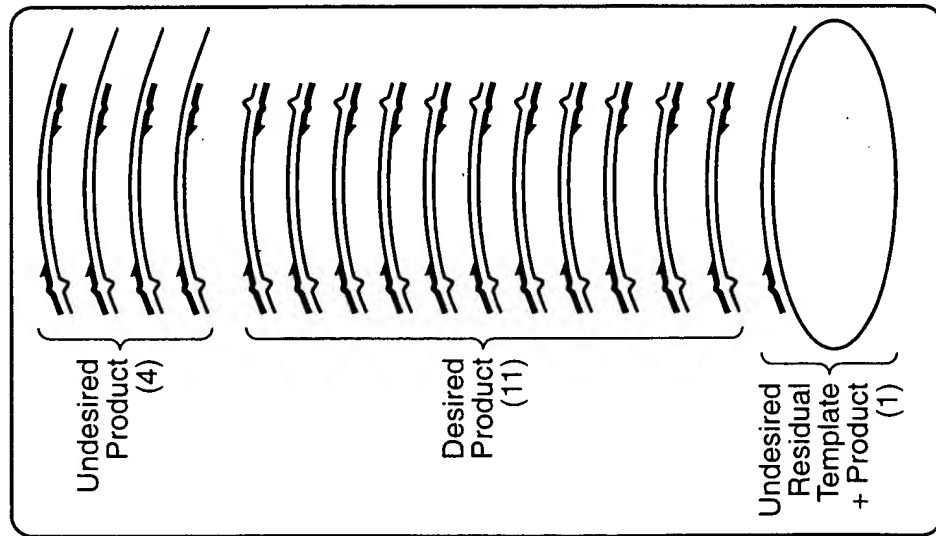


FIG.--9B